ELECTROPHORETIC APPARATUS FOR THE QUANTITATIVE ELUTION OF PROTEINS OR POLYPEPTIDES FROM A GEL

BACKGROUND OF THE INVENTION

The present invention relates to an apparatus for the quantitative elution of proteins or polypeptides from a gel by means of elctrophoresis.

Protein mixtures, such as viruses, and isolated biological membranes, etc. (separated according to SDS (sodium dodecyl sulfate)/polyacrylamide gel electrophoresis (see e.g. Laemmli, Nature Vol. 227, pp. 680 to 685, 1970; and Thomas and Kornberg, Proc. Nat. Acad. Sci. U.S.A. Vol. 72, pp. 2226 to 2630, 1975) are represented as individual bands in a plate gel. These bands correspond to individual proteins having the corresponding molecular weight. In order to utilize the thus separated proteins, or polypeptides, for other purposes, the bands are separated from the plate gel and collected. The protein retained in the network of the polyacrylamide is electrophoretically eluted. Generally a running buffer such as 5 mM tris-glycin, pH 8.6 0,2% SDS and 0.5% mercaptoethanol is used as the elution buffer.

SUMMARY OF THE INVENTION

It is an object of the present invention to provide an apparatus with which isolation of even larger quantities of electrophoretically separated proteins or polypeptides is possible in a desired concentration.

The above and other objects are accomplished in accordance with the invention wherein an apparatus is provided for the quantitative elution of proteins or polypeptides from a gel by means of electrophoresis. The apparatus includes an upper electrode chamber for holding a buffer solution containing the gel from which the proteins or polypeptides are to be eluted. An upper electrode is mounted in the upper electrode chamber. A lower electrode chamber for holding buffer solution is disposed beneath the upper electrode chamber. A lower electrode is mounted in the lower electrode chamber. A septum separates the upper electrode chamber from the lower electrode chamber. The septum is provided with a connecting passage for connecting the upper and lower chambers. A collecting capsule for collecting the proteins or polypeptides is disposed at the end of the connecting passage in the lower electrode chamber, and is adapted to be suspended in the buffer solution which is to be held in the lower chamber.

In operating the apparatus, the application of a voltage potential between the upper and lower electrodes causes the proteins or polypeptides to be eluted from the gel in the upper chamber and collected in the collecting capsule.

It is to be understood that both the foregoing general description and the following detailed description are exemplary, but are not restrictive of the invention.

BRIEF DESCRIPTION OF THE DRAWING

The sole FIGURE is a cross-sectional view of an elution apparatus according to the invention.

DESCRIPTION OF THE PREFERRED EMBODIMENT

Referring now to the drawing, there is shown an elution apparatus which includes an upper electrode chamber **4** which is to contain a buffer solution and a lower electrode chamber **10** which is to contain the same buffer solution. The individual proteins or polypeptides separated and collected from the plate gel which are to be eluted, are placed in the buffer solution in upper electrode chamber **4**.

Chambers **4** and **10**, both of which can be of cylindrical design, are placed one on top of the other and are separated from each other by a septum **30**. Septum **30** is provided with a connecting passage **13** which connects upper electrode chamber **4** with lower electrode chamber **10**.

Chamber **4** is formed by a side wall **32**, a bottom **16** and a removable top **3**. Bottom **16** generally comprises the upper surface of septum **30** which preferably is integral (in one piece) with side wall **32**. A negative electrode **1** is provided in top **3**.

Chamber **10** is formed by a side wall **34**, a bottom **12** and a top **36** which is the lower surface of septum **30**. A positive electrode **11** is provided in bottom **12** of chamber **10**.

Electrodes **1** and **11** are illustrated with a planar shape, but could have an annular or other suitable shape. Both electrodes **1** and **11** are disposed perpendicular to the longitudinal axis (axis of rotation) of chambers **4** and **10**, which also passes through connecting passage **13**.

Upper electrode **1** is fastened to a mount **14** which passes through top **3**. A guide ring **15** is attached to the upper surface of top **3** and is provided with a thumb screw **2**. Mount **14** passes through guide ring **15** and can be set at any desired height by adjusting thumb screw **2**. Thus, upper electrode **1** can be adjusted in height with respect to the surface (not shown) of the gel-buffer solution disposed in upper electrode chamber **4**. The height at which electrode **1** is set depends on the fill level of the gel-buffer solution in chamber **1** and the electrical voltage between electrodes **1** and **11**. The level of the elution buffer in the upper electrode chamber **4** should be 1.5 to 2 cm below the upper rim of the chamber. The electrode **1** of the upper chamber should dipp about 1 cm into the buffer solution. The lower electrode chamber **10** should be filled with buffer up to the level of the counter ring **8**.

The electrical voltage between the electrodes **1** and **11** should not exceed 100 V.

Bottom **16** of upper electrode chamber **4** is preferably constructed in the form of a funnel having a downwardly narrowing conical inclined surface **38**, a funnel neck **40** which meets with inclined surface **38**, and a seat **42** which meets with funnel neck **40**. The lowest point of bottom **16** is at seat **42** and is joined to connecting passage **13**. A small tube **7** having an upper end **44** and a lower end **19** is disposed in passage **13** and extends into the area defined by funnel neck **40** and seat **42**. Tube **7** is provided with an abutment **17** which meets a corresponding abutment **46** in passage **13**. Abutments **17** and **46** together define the extent to which tube **7** can be inserted from lower electrode chamber **10** into connecting passage **13** and the area defined by neck **40** and seat **42**. A radial packing ring **6** is provided at neck **40** and seals tube **7** relative to upper electrode chamber **4**. Sinter plates **5** may cover upper end **44** and lower end **19** of tube **7**. The radial packing ring **6** is a oil seal or rubber O-ring. Glass sinter plates **5** of 150 μm to 50 μm pore sizes can be used.

Lower end **19** of tube **7** has a frustoconical shape. A dialysis capsule **9** is clamped onto end **19** by a counter cone ring **8**.